ROLE OF GENETIC FACTORS AND FOWL POX VIRUS IN PARTHENOGENESIS IN TURKEY EGGS

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THREE live poultry viruses—fowl pox, Rous sarcoma and Newcastle disease—have been employed experimentally at the Agricultural Research Center, Beltsville, Maryland, in studies designed to test their action on parthenogenesis in eggs of Beltsville Small White (BSW) turkeys. Each of the viruses was found to have an enhancing influence on cells of parthenogenetic origin (OLSEN 1956, 1961; OLSEN and POOLE 1962 and unpublished observations). The same viruses, however, on being inactivated with beta-propriolactone and subsequently used to inoculate virgin BSW turkeys, had no affect on the level of macroscopic observable parthenogenesis in unfertilized eggs (OLSEN 1962).

Selective breeding has likewise been shown to be an effective method of increasing the predisposition for parthenogenesis in BSW turkeys. Between 1952 and 1963, the average level of parthenogenesis in turkey eggs at this station increased from 16.7% to over 40%. The most significant increase came in numbers of embryos, from a level of 0.2% in 1952 to about 12% in 1963 (OLSEN 1965). All turkeys were routinely vaccinated each year for fowl pox on reaching 6 to 8 weeks of age and again at 30 to 32 weeks.

The relative importance of roles played by genetic factors and fowl pox virus, especially in embryo production and hatched parthenogens, remains obscure. It was most desirable, therefore, that a series of breeding tests be initiated whereby two genetically similar populations of turkeys would be selected for a high and/or low incidence of parthenogenesis, one group of birds in the presence of fowl pox virus, the other in the presumed absence of this virus.

A cooperative study designed to obtain information on the respective roles of genetic factors and fowl pox virus in parthenogenesis was initiated in 1961 with Pennsylvania State University, University Park, Pennsylvania.

MATERIALS AND METHODS

Thirty-two mature, nonvaccinated turkey females were furnished by Pennsylvania State University. This group of pedigreed birds consisted of 16 full-sister pairs of Pozo Gray (PG) turkeys. This strain was selected because it had been identified as having a low incidence of parthenogenesis, 1 to 2%. One member of each pair of full sisters (16 birds) was brought to

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TABLE 1

Incidence of parthenogenesis for the original 16 pairs of Pozo Gray turkeys reared at the Pennsylvania State University, University Park

Beltsville				University Parks			
Wingband	Eggs set No.	Parthenogenesis No.	%	Wingband	Eggs set No.	Parthenogenesis No.	%
D 21 E	50	2	4.0	D 21 N	24	0	0.0
D 21 G	45	2	4.4	D 21 Q	26	0	0.0
D 21 H	52	0	0.0	D 21 U	23	0	0.0
D 21 V	41	2	4.9	D 21 X	24	0	0.0
D 22 E*	53	8	15.1	D 22 D*	32	1	3.1
D 22 J	43	0	0.0	D 22 P	33	1	3.0
D 23 P	21	2	9.5	D 23 H	31	0	0.0
D 25 F	70	0	0.0	D 25 J	30	0	0.0
D 25 L	51	0	0.0	D 25 K	30	0	0.0
D 25 O	61	1	1.6	D 25 Q	31	1	3.2
D 28 C	61	1	1.6	D 28 G	36	0	0.0
D 28 J	62	1	1.6	D 28 H	34	1	2.9
D 29 L	45	1	2.2	D 29 F	32	0	0.0
D 29 N*	44	5	11.4	D 29 G*	33	1	3.0
D 29 O*	46	6	13.0	D 29 H	28	0	0.0
D 30 L	35	0	0.0	D 30 J	27	0	0.0
	780	31	4.0		474	5	1.1

* Progeny selected for the next generation.

Beltsville when the turkeys were 24 weeks old and vaccinated for fowl pox. The other 16 birds were retained at PSU to serve as nonvaccinated controls.

Wingband numbers of the 16 original paris are listed in Table 1. Lettering on each band shows family relationships, the whole number indicating the dam, the first letter the year and the last letter the individual bird. The distribution of stocks, and numbers and percentages of parthenogenetic eggs for each of the hens are also shown in Table 1.

Housing and management conditions were standardized as far as practical so as to minimize, at each station, environmental influences which might conceivably affect the level of parthenogenesis. Turkeys on test were kept in cages at Beltsville in all years of the experiment. At University Park, the turkeys were kept in cages during the first year, after which time they were confined in floor pens. Birds at both locations received 14 hours of light daily, starting on January 1 each year, when the virgin females were 28 to 32 weeks of age.

Eggs, as collected, were identified by hen numbers and date of lay. Each evening they were placed in forced-draft incubators operating at temperatures of 37.5°C and at a relative humidity of 57%. Nearly all eggs were incubated for 10 days before being broken into water, where their blastodiscs were examined macroscopically for parthenogenetic development. Occasionally, however, some eggs were examined either on the 9th or 11th day of incubation. Eggs whose germinal discs measured 6 mm or more in diameter were classified as having undergone parthenogenetic development; if no increase was noted in size of the disc, they were listed as nonparthenogenetic.

Several exchange visits were made by both investigators each year during the 60–90 day testing period. On such occasions, sample lots of incubated, unfertilized eggs were broken and classified for parthenogenetic development. These visits thus helped to insure the maintenance of a uniform system of classification of parthenogenetic development.

Each year, following completion of parthenogenetic tests, certain virgin hens were mated to

TABLE 2

Number of Pozo Gray females tested and the number of hens selected as breeders each year. Also shown are average percentages of parthenogenesis and the number of males used

	Year of testing									
	1962		1963		1964		1965		1966	
	No.	% Partheno-genesis	No.	% Partheno-genesis	No.	% Partheno-genesis	No.	% Partheno-genesis	No.	% Partheno-genesis
ARC-H* (Beltsville)										
Females tested	16	4.0	17	7.1	38	8.5	24	16.6	24	21.1
Females selected	3	13.3	7	11.6	8	20.0	12	21.2	..	...
Males used	2	...	7	...	3	...	4	...	..	..
PSU-H† (University Park)										
Females tested	16	1.1	13	5.2	33	7.0	37	8.6	22	18.6
Females selected	2	3.1	7	7.4	9	18.7	11	14.2	..	...
Males used	2	...	5	...	3	...	3	...	..	..
PSU-L† (University Park)										
Females tested	16	1.1	14	1.2	37	1.1	54	0.3	18	1.0
Females selected	5	0.8	8	0.0	12	0.0	10	0.5	..	...
Males used	3	...	4	...	2	...	2	...	..	...

* Given five fowl pox vaccine at approximately 6–8 and again at 30–32 weeks of age.
† Not vaccinated.

obtain stock for the following season. Selection of breeding stock each year was under the supervision of the second author and was based on records compiled and stocks available at both Beltsville and University Park. Matings were always made with the objective of reproducing and maintaining at each location, stocks stemming from identical families. Hens selected each year as breeders for the high incidence lines were from families showing the most marked predisposition for parthenogenesis. At University Park, a line was also selected for a lower than average incidence of parthenogenesis. Males, likewise, were sibs of the females selected from families having the highest or lowest average incidence of parthenogenesis. The numbers of females tested and selected each year and the numbers of males used are shown in Table 2.

The same two males serving as sires at University Park in 1962 were also used that same year as sires at Beltsville. After the two males were brought to Beltsville, they were vaccinated for fowl pox before being used as breeders. Each male was mated to both sisters of selected families, one sister at each station. Thereafter, males came from stocks hatched, raised, and maintained at each of the respective stations. Starting in the second year, all matings were brother and sister.

Poults at each station were brooded on the floor. Those raised at Beltsville were vaccinated twice each season for fowl pox, the first upon reaching 6 to 8 weeks of age, the second at 30 to 32 weeks. Poults hatched and raised at University Park were not vaccinated for fowl pox. There was no evidence that fowl pox was ever present in the turkeys at University Park during the course of the experiment.

In 1965 all eggs from the two stations were incubated at University Park. Upon hatching, poults from each dam were divided, one half being sent to Beltsville and the remainder kept at University Park. Eggs from vaccinated stock at Beltsville were tested at University Park in the absence of fowl pox virus, while eggs of their full sisters were tested at Beltsville following vaccination. Likewise, pairs of sisters from high and low lines developed at University Park were tested at both stations. Thus, the stock tested at each station from each line was as much alike genetically, as possible.

RESULTS

At the start of the experiment in 1962, 11 of 16 hens at Beltsville produced one or more eggs exhibiting development. The incidence of parthenogenetic eggs ranged from 0 to 15.1%. Five of 16 hens at University Park produced eggs showing development, with variations of 0 to 3.8% parthenogenesis.

Table 3 gives total numbers of unfertilized eggs incubated, and numbers and percentage of these eggs showing parthenogenetic development. The eggs were from three categories of turkeys: PSU (University Park) high incidence strain, PSU low incidence strain, and the ARC (Beltsville) high incidence strain.

Percentage values given in the last column of Table 3 show the overall incidence of parthenogenesis encountered each year in eggs from each line, and the effectiveness of selective breeding (with and without the employment of fowl pox virus) in either increasing or decreasing the level of parthenogenesis. At Beltsville, the average level of parthenogenetic development increased from 4.0% in 1962 to 21.0% in 1966. At University Park, parthenogenetic development in the high incidence line increased from a level of 1.1% in 1962 to 18.6% in 1966. No change was noted in the average level of parthenogenesis in the low incidence

TABLE 3

Incidence of parthenogenetic development in unfertilized eggs of young, virgin Pozo Gray turkeys

Year	Testing station	Line of turkeys	No. hens	No. eggs tested	Number and percentages of eggs found to contain: Membrane only No.	Membrane only %	Blood and membrane No.	Blood and membrane %	Embryos No.	Embryos %	Total parthenogenesis No.	Total parthenogenesis %
1962	ARC*	ARC	16	780	30	3.9	0	0.0	1	0.1	31	4.0
	PSU†	PSU	16	474	5	1.1	0	0.0	0	0.0	5	1.1
1963	ARC	ARC-H‡	17	535	36	6.7	0	0.0	2	0.4	38	7.1
	PSU	PSU-H	13	153	8	5.2	0	0.0	0	0.0	8	5.2
	PSU	PSU-L	14	167	2	1.2	0	0.0	0	0.0	2	1.2
1964	ARC	ARC-H	38	870	68	7.8	4	0.5	2	0.2	74	8.5
	PSU	PSU-H	33	931	63	6.8	2	0.2	0	0.0	65	7.0
	PSU	PSU-L	37	1217	14	1.1	0	0.0	0	0.0	14	1.1
1965	ARC	ARC-H	24	975	145	14.9	12	1.2	5	0.5	162	16.6
	PSU	PSU-H	37	1234	104	8.4	3	0.2	0	0.0	107	8.6
	PSU	PSU-L	54	2077	7	0.4	0	0.0	0	0.0	7	0.4
1966	ARC	ARC-H	24	1022	196	19.1	16	1.6	4	0.4	216	21.1
	PSU	ARC-H	24	986	155	15.7	28	2.8	5	0.5	188	19.1
	ARC	PSU-H	22	866	161	18.6	10	1.2	3	0.3	174	20.1
	PSU	PSU-H	22	627	100	15.9	17	2.7	0	0.0	117	18.6
	ARC	PSU-L	18	625	17	2.8	1	0.1	0	0.0	18	2.9
	PSU	PSU-L	18	593	6	1.0	0	0.0	0	0.0	6	1.0

* ARC—Agricultural Research Center, USDA, Beltsville, Maryland.
† PSU—The Pennsylvania State University, University Park, Pennsylvania.
‡ H—High Incidence, L—Low Incidence.

strain at University Park following four years of selection against parthenogenesis.

Evidence of blood formation was found in 32 of 3,402 eggs (0.94%) from the turkeys selected and tested at Beltsville for a high incidence of parthenogenesis. Blood formation was encountered in 22 of 2,945 eggs (0.75%) from the line selected and tested for increased incidence at University Park, and in none of 4,054 eggs from the low incidence line during the years 1963 through 1966.

A total of 22 parthenogenetic embryos was encountered. Seventeen of the 22 embryos were found in eggs of vaccinated birds tested at Beltsville. The other five embryos were found at University Park in 1966 in eggs of progeny coming from the line of vaccinated Grays developed at Beltsville. No recognizable parthenogenetic embryos were encountered at University Park in eggs of nonvaccinated females whose dams and sires had not been vaccinated for fowl pox.

DISCUSSION

Our results indicate that both genetic factors and live fowl pox virus play an active role in parthenogenetic development. Genetic influences are clearly in evidence, since the average level of parthenogenesis in eggs of Pozo Gray (PG) turkeys at University Park (unorganized growth) increased through selection from a level of 1.1% in 1962 to 18.6% in 1966, even in the absence of fowl pox virus. Furthermore, these selection effects were manifested in a relatively short period of time, i.e., after only four years of selection. Advances in the incidence of parthenogenetic development have been attained in Beltsville Small Whites (BSW) at Beltsville through selection of both males and females. These results support those of OLSEN (1965) who reported an increase in parthenogenesis from a level of 16.7% to over 40% in a 9-year period. It should be pointed out, however, that selection of BSW turkeys was done in the presence of fowl pox virus. Selection responses with PG turkeys are in agreement with those obtained with Drosophila. STALKER (1954), working with *Drosophila parthenogenetica* was able through selection over a period of 17 generations to raise the production of viable progeny from 8/10,000 to 151/10,000 unfertilized eggs. Selection improvement in parthenogenesis occurred during the first 17 generations and thereafter ceased. He likewise succeeded in increasing the level of parthenogenesis in eggs of *Drosophila polymorpha* from 1/10,000 to 133/10,000 (STALKER 1956).

CARSON (1962), working with two crossbred strains of Drosophila, was able in 12 generations of selection, to increase the incidence of parthenogenesis approximately 34-fold over the rate in unselected stocks. The same investigator (1967) working with eggs of several strains of *Drosophila mercatorum* succeeded in raising the level from about 1% to 6.0% in the course of 130 generations. CARSON found that both males and females transmitted the parthenogenetic trait to their offspring. He further states that the predisposition for parthenogenesis is influenced by a number of independent loci.

A total of 22 PG parthenogenetic embryos were encountered during this 5-year study, in eggs of vaccinated stock at Beltsville or in eggs produced in 1966 at

University Park by turkeys originating from eggs transferred from Beltsville in 1965. At Beltsville, one or more embryos were found each year in eggs of vaccinated stock. Four years of intensive selection for an increased incidence of parthenogenesis at University Park, however, failed to give rise to a single hen with the ability of producing eggs capable of forming true embryos. This was so in spite of a marked increase at University Park in the overall average percentage increase of unorganized parthenogenetic development (16-fold) in five years. Whether true embryo production eventually will be achieved in unfertilized eggs of nonvaccinated PG turkeys at University Park is still an unanswered question.

The nature of the action of fowl pox virus on unfertilized turkey ova has not been clearly established. It has been shown, however, that following employment of this DNA virus, the percentage incidence of parthenogenesis in eggs of the same hens increases markedly over the level recorded for their eggs before time of vaccination (OLSEN 1956). Most of the increase comes in the number of live embryos, suggesting that the virus may be serving as an organizer as well as a stimultant to cellular proliferation. The effect of the virus seems to persist in eggs of daughters and granddaughters even if subsequent generations of birds are not vaccinated. The prolonged effect suggests that the inheritance of such birds may be altered in some manner by the virus, enabling certain hens to produce unfertilized ova, some of which have a strong predisposition for parthengenesis. It would be interesting to know if live viruses, especially the DNA variety, have an influence on embryo production in species of insects which reproduce by parthenogenesis.

SUMMARY

Two closely related groups of Pozo Gray turkeys were compared; one group vaccinated with live fowl pox virus, was maintained at Beltsville, the other, unvaccinated, at University Park. This 5-year study showed that (1) parthenogenesis (unorganized type) increased in Pozo Grays through selection without fowl pox virus, (2) all parthenogenetic embryos from Pozo Gray turkeys appeared in eggs of vaccinated stock or in eggs of progeny of vaccinated birds.

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